A Rough Guide to S-DIVA

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Contents

A Rough Guide to S-DIVA 1

1. Introduction (back to contents) 3

1.1 Explanation 3

1.2 Citation 3

2. Installing (back to contents) 4

3. A simple S-DIVA tour step by step (back to contents) 5

3.1 Preparation 5

3.2 Load Input Files 5

3.3 Analysis 6

3.4 Results 7

4. How to …(back to contents) 9

4.1 How to make Trees data set. 9

4.2 How to make a Final tree. 10

4.3 How to make Distributions file 11

4.4 Examples 11

5. Explanation of messages (back to contents) 13

6. References (back to contents) 15

1. Introduction (back to contents)

1.1 Explanation

Studies in historical biogeography based on phylogeny have accumulated rapidly in the literature due to the exponential increase in phylogenetic studies (see Xiang et al., 1998, 2004, 2005, 2006; Wen, 1999; Sanmartín et al., 2001; Donoghue & Smith, 2004; Sanmartín & Ronquist, 2004; Soltis et al., 2006; Harris et al., 2009). Dispersal–Vicariance Analysis (DIVA) (Ronquist, 1997, 2001) is one of the most widely used methods of inferring biogeographic histories. This is evidenced by an advanced Google scholar search for "DIVA" and "biogeography" in 2009 returning greater than 50 relevant results. DIVA reconstructs the ancestral distribution in a phylogeny by optimizing a three-dimensional cost matrix, in which extinctions and dispersals "cost" more than vicariance (Ronquist, 1997; Lamm & Redelings, 2009). One problem with the current implementation of DIVA is that it ignores the uncertainty in phylogenetic inference because ancestral ranges are reconstructed onto a fixed tree topology assumed to be without error (Nylander et al., 2008). A second source of uncertainty in DIVA is that associated with ancestral area optimization; multiple equally optimal reconstructions often result in multiple ranges suggested at ancestral nodes (Ronquist, 1997, Nylander et al., 2008).

To account for these uncertainties, Nylander et al. (2008) and Antonellia A et al.(2009) recently showed the utility of a non-parametric empirical Bayesian approach to DIVA. Their approach handles phylogenetic uncertainty and uncertainty in DIVA optimization. Harris & Xiang (2009) proposed an alternative approach to Bayes-DIVA, which differs in its ability to handle uncertainty at some nodes.

We have written Statistical Dispersal-Vicariance Analysis (S-DIVA), a program which complements DIVA, implements the methods of Nylander et al. (2008) and Harris and Xiang (2009), and determines statistical support for ancestral range reconstructions using a novel method, the S-DIVA value. In S-DIVA, the frequencies of an ancestral range at a node in ancestral reconstructions are averaged over all trees and each alternative ancestral range at a node is weighted by the frequency of the node occurring or by some other measure of support for the node. S-DIVA is easy-to-install, provides a user-friendly graphical interface, and generates exportable, graphical results.

1.2 Citation

Program:

Yu Y, AJ Harris, X He. S-DIVA (Statistical Dispersal-Vicariance Analysis) [VERSION]. Available at http://mnh.scu.edu.cn/S-DIVA

Associated Materials:

Yu Y, Harris AJ, He XJ. 2010. S-DIVA (Statistical Dispersal-Vicariance Analysis): a tool for inferring biogeographic histories. Molecular Phylogenetics and Evolution. doi:10.1016/j.ympev.2010.04.011

Ronquist, F. (2001) DIVA version 1.2. Computer program for MacOS and Win32. Evolutionary Biology Centre, Uppsala University. Available at http://www.ebc.uu.se/systzoo/research/diva/diva.html.

User's Guide:

Yu Y, AJ Harris, and X He. (2010). A rough guide to S-DIVA. Available online at http://mnh.scu.edu.cn/S-DIVA. Accessed [DATE].

2. Installing (back to contents)

S-DIVA was developed for use on Microsoft Windows® operating systems:

Supported Operating Systems: Windows NT, 2000, 2003, 2008, XP, Vista and Windows 7

Processor: 400 MHz Pentium processor or equivalent (Minimum); 1GHz Pentium processor or equivalent (Recommended)

RAM: 64 MB (Minimum); 128 MB (Recommended)

Display: 800 x 600, 256 colors (Minimum); 1024 x 768 high color, 32-bit (Recommended)

For computation of large datasets, you should have a fast processor and large amount of free space on disk.

(*Note: You can use [**File > Clean Cache**] in S-DIVA to free the disk space after the analysis.*)

S-DIVA is distributed without charge by download from the S-DIVA web site, http://mnh.scu.edu.cn/S-DIVA. If someone has given you a copy of S-DIVA, we strongly suggest that you download the most recent version from this site. No installation is needed and you can put the S-DIVA.exe in any directory you like.

DIVA 1.2 (Ronquist, 1997, 2001) is bundled in S-DIVA. You can also download it from here and put it in the directory of S-DIVA.

IMPORTANT: If you are using Windows 2000, 2003 or XP, please make sure that Microsoft® .NET 2 Framework is installed on your computer. This is usually installed through Windows updates, but it may be absent from older systems. The .NET framework 2 packages are available for free here and should be installed prior to using S-DIVA.

For Mac and Linux Users

The S-DIVA web service (http://mnh.scu.edu.cn/sdiva_web) is a good choice for MAC and Linux users. We have added a note about the availability of the web service in the Implementation Details. Parallels Desktop or VMware Fusion can also be used to run S-DIVA on a Mac or Linux. The S-DIVA web service is still in beta and we recommend the offline version of S-DIVA.

3. A simple S-DIVA tour step by step (back to contents)

3.1 Preparation

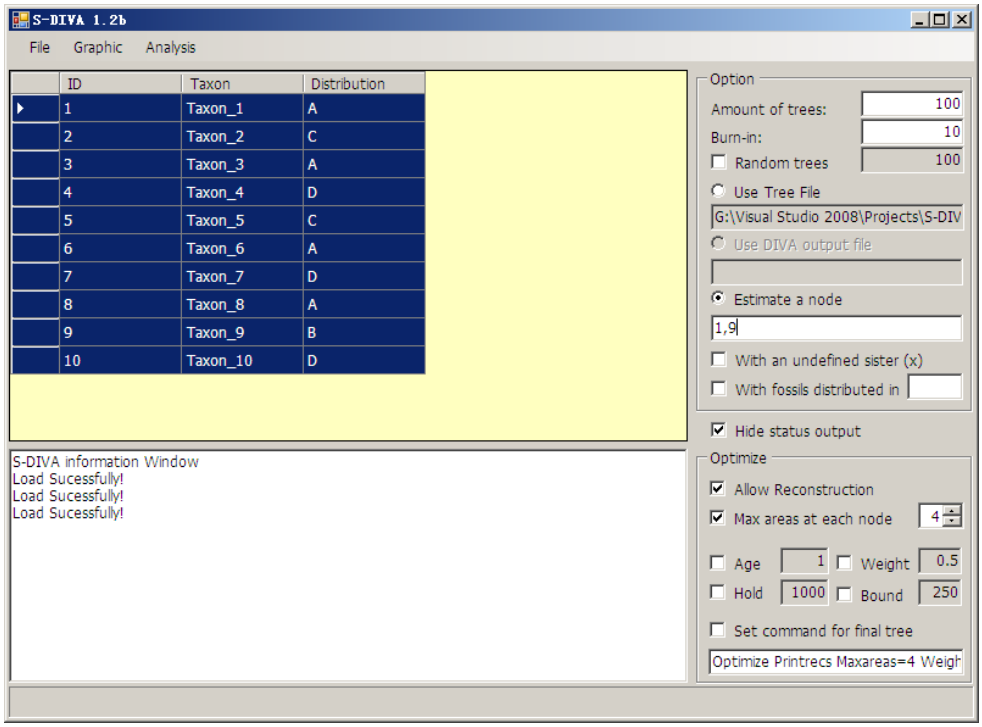
You need:

1. **Trees data set.** (Output from BEAST (Drummond and Rambaut, 2006) or PAUP* (Swofford, 2003))
   (A sample trees data file: "sample\sample1.tree")
   -[How to]: How to obtain trees data set from BEAST.
   -[How to]: How to obtain trees data set from PAUP.
   -[How to]: How to obtain trees data set from other phylogenetic programs.
2. **A Final tree (not required if you want to estimate a particular node only).** (Output from BEAST or PAUP*)
   (A sample final tree: "sample\Sample1_Final_Tree.tre")
   -[How to]: How to make a final tree using BEAST.
   -[How to]: How to make a final tree using PAUP.
   -[How to]: How to obtain a final tree from other phylogenetic programs.
3. **Distributions file (not required).**
   (A sample distributions file: "sample\Sample1_Distribution.csv")
   -[How to]: How to make a Distributions file.
   -[How to]: How to input distributions in S-DIVA.

3.2 Load Input Files

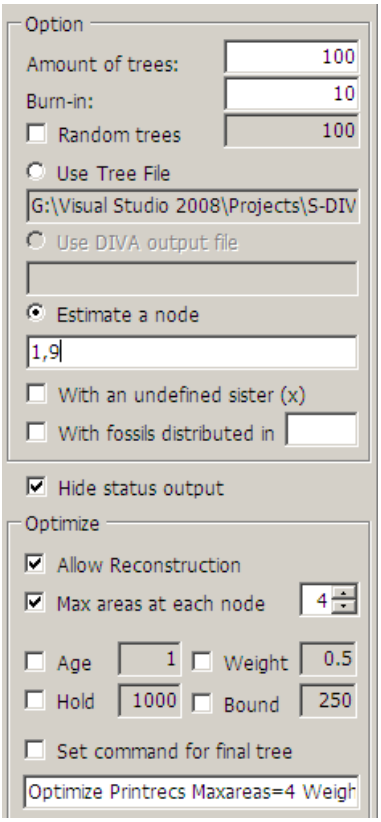
Launch S-DIVA

1. Open [**File > Load Trees**] and navigate to your trees data set and select it.
   [Example: Open "Sample1.trees" in folder Sample]
2. Open [**File > Load Computed File > Load Final Tree**] and navigate to your final tree file and select it.
   [Example: Open "Sample1_Final_Tree.tre" in folder Sample]
3. If you have a distributions file then Open [**File > Load Distribution**] and navigate to your file and select it.
   [Example: Open "Sample1_Distribution.csv" in folder Sample]
   You can input and revise the distributions in the entry fields in the Distribution column in S-DIVA.



3.3 Analysis

Choose [**Analysis > Run Analysis**] to use default options to run analysis.



[Options]

Amount of trees: The total number of trees in your trees data set. You can decrease this number to discard the trees from the end of the trees data set.

Burn-in: The number of trees that will be discarded from the beginning of the trees data set. [Example: ***Amount of trees=10001*** and ***Burn-in*=1000** means: trees from 1001 to 10001 in trees data set will be included in next analysis and all others will be ignored]

Use Tree File: Run S-DIVA analysis for all nodes in the final tree.

Use DIVA output file: Allows you to use an existing DIVA output file as the result of the final tree. (A sample DIVA output file: Sample\Sample1_DIVA_output.txt)

You may use the following command to get an output file form DIVA:

```
echo status;
output out.txt;
tree ((((5,(((2,1),3),4)),(((7,6),8),9)),10),11); (replace with your tree)
distribution A A A AB B B B BC A ABC C; (replace with your distribution)
Optimize Printrecs;
quit;
```

Estimate a particular node: Run S-DIVA only for a particular node. You can specify a node using the following format:

1,2,3,4,5 or 1-5 1,2,3,4,8,9,10 or 1-4,8-10 (Numbers correspond to **ID**s in the Distributions column in the S-DIVA interface.)

With an undefined sister (x): Specify a clade by the node shared with its sister and estimate the ancestral range of the clade and an undefined sister (*x*) (Harris & Xiang, 2009). Specify the node using one of the formats shown above.

With omitted taxon distributed in: Add distribution information for and topological placement of a taxon omitted from the phylogeny. S-DIVA manually places the unnamed taxon as sister to the specified node and considers its distribution when running the analysis. Note: The manually placed taxon is not included in graphical output.

Random tree: Select random trees form trees data set to run the analysis. Trees will be selected from between ***Burn-in*** and ***Amount of trees***. You can save the randomly selected trees to a new file with [**File> Save Processed Trees**].

Hide detailed output: Hide the output of analysis. You can change this option while running.

[Optimize]

Allow Reconstruction: Unchecking this option changes the method used for calculating $F(x_n)$ from i/D_t to $1/N$. See section 3.4.1 below.

Max areas: The number of unit areas allowed in ancestral distributions. **Weight, Age, Bound and Hold**: These four options are the same as in DIVA 1.2.

Set command for final tree: Set the optimize command for the final tree separately.

If you need additional help with the [**Optimize**] options ***Max areas***. ***Weight***, ***Age***, ***Bound***, ***Hold***, or with setting the command line, we recommend reviewing these options in DIVA 1.2 (command: help;) and the DIVA 1.1 User's Manual (Ronquist, 1996).

3.4 Results

3.4.1. Text results:

If **Allow Reconstruction** is checked**,** you will see results in the **S-DIVA information Window that look** like this:

[

****

>>>

3.4.2 Graphic results

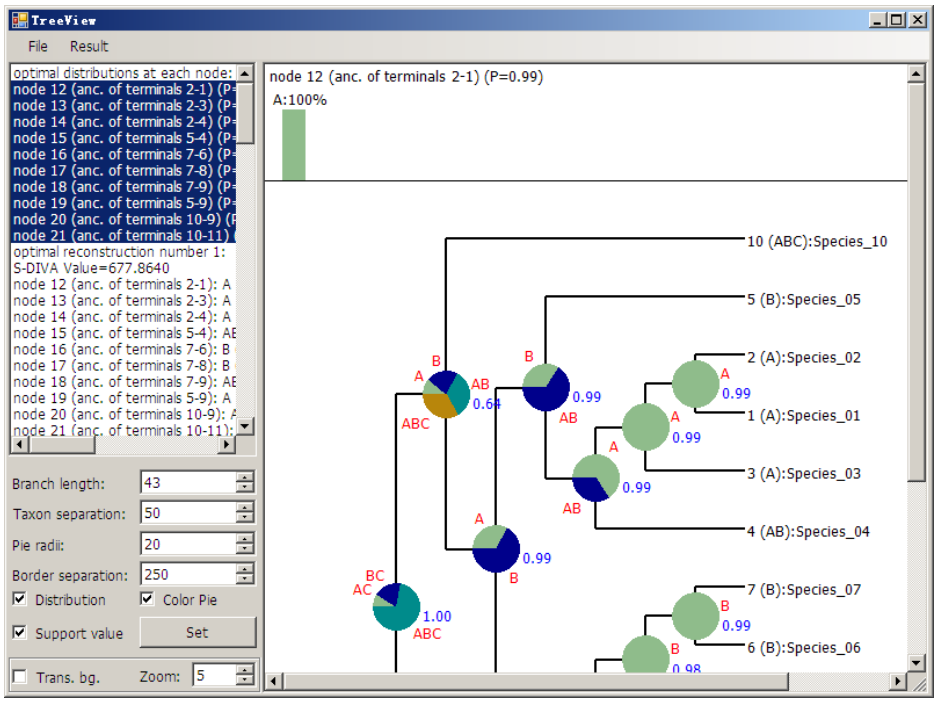
After running an analysis, a ***Tree View*** window will automatically open. You can also access this window from [**Graphic > Tree View**].

Node list:

[**optimal distributions at each node**]: Select a node to display alternative ancestral distributions (pie chart and bar chart) at the selected node on the tree. Select all nodes in the list to display pie charts and distributions for all nodes as in the figure below. Bar charts are displayed for only one node at a time.

[**optimal reconstruction number <*n*>**]: Select all nodes to display distributions at all nodes from reconstruction <*n*> on the tree. Select a single node to display the distribution at that node from reconstruction <*n*> on the tree.

Display options:


TreeView
File
Result
optimal distributions at each node:
node 12 (anc. of terminals 2-1) (P=
node 13 (anc. of terminals 2-3) (P=
node 14 (anc. of terminals 2-4) (P=
node 15 (anc. of terminals 5-4) (P=
node 16 (anc. of terminals 7-6) (P=
node 17 (anc. of terminals 7-8) (P=
node 18 (anc. of terminals 7-9) (P=
node 19 (anc. of terminals 5-9) (P=
node 20 (anc. of terminals 10-9) (P
node 21 (anc. of terminals 10-11) (
optimal reconstruction number 1:
S-DIVA Value=677.8640
node 12 (anc. of terminals 2-1): A
node 13 (anc. of terminals 2-3): A
node 14 (anc. of terminals 2-4): A
node 15 (anc. of terminals 5-4): AB
node 16 (anc. of terminals 7-6): B
node 17 (anc. of terminals 7-8): B
node 18 (anc. of terminals 7-9): AB
node 19 (anc. of terminals 5-9): A
node 20 (anc. of terminals 10-9): A
node 21 (anc. of terminals 10-11):
Branch length: 43
Taxon separation: 50
Pie radii: 20
Border separation: 250
Distribution
Color Pie
Support value
Set
Trans. bg.
Zoom: 5
node 12 (anc. of terminals 2-1) (P=0.99)
A:100%
10 (ABC):Species_10
5 (B):Species_05
2 (A):Species_02
1 (A):Species_01
3 (A):Species_03
4 (AB):Species_04
7 (B):Species_07
6 (B):Species_06
0.99
0.64
1.00
0.98

The node of terminals 2 (species_02) to 3(species_03) seems to have originated in area A with 0.99 support value. The node of terminals 10 (species_10) to 9 (species_09) have four possible ancestral ranges "A, B, AB or ABC", the occurrence of each range is A: 11%, B: 23%, AB: 33%, ABC: 33%. The selected node's distributions (node 20, terminals 10-9) are displayed as bar chart.

Branch length: Set the length of branches (10-1000px). All branches in the cladogram become proportionately larger or smaller.

Taxon separation: Set the separation of taxa (10-200)

Pie radii: Set the radii of pie charts (2-100)

Border separation: set the width of the white space in the tree view window (10-2000). Note that it is possible to make this too small for the width of the tree.

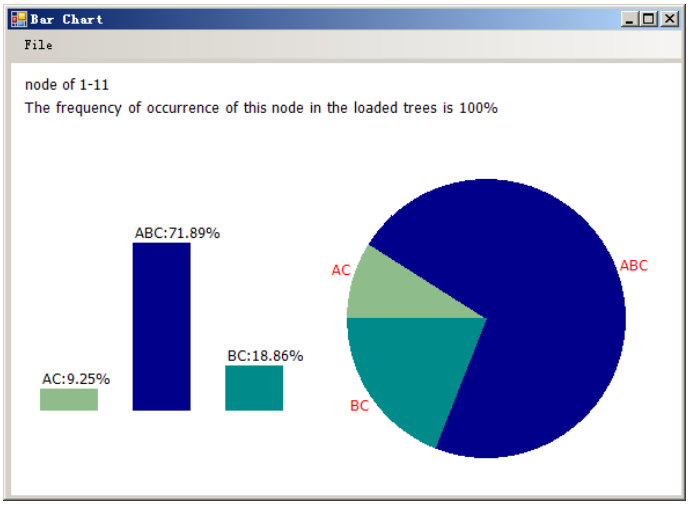
Distribution: Show distribution areas. These will display as red letters next to the pie charts.

Color Pie: Show colored pie chart. Unchecking this will result in grayscale pie charts.

Support value: Show support value for the node in the sample of trees analyzed (i.e., p_n)

Options for save picture:

Trans. bg.: save the picture with transparent background. If unchecked, the picture will be saved with a white background.



Zoom: The measure of magnification of saved picture.

Mouse control:

[*Double click* on a node]: Select the node.

[*Right click* on a node]: Show or hide the pie chart

[*Left click* on a node]: Show the bar chart.

The left figure shows a result for estimating a particular node. This can be accessed from from [**Graphic > Node View**].

4. How to ...(back to contents)

In the compressed ZIP file download from here, you will find a "sample" folder. All sample files referenced in this manual are in the sample folder.

4.1 How to make Trees data set.

4.1.1 How to obtain trees data set from BEAST. (Back to Top)

- Launch ***BEAUti v*.exe***
- Select **File > Import Alignment** and navigate to your NEXUS input file.
  (From our sample files, we would select *sample1.nex* in the Sample folder)
  (Note: If ***BEAUti*** can't load the NEXUS file then load the file into PAUP and export it use "**format=NEXUS**" option.)
- Select MCMC panel, set the number of generations the MCMC algorithm will run for.
  (We set **length of chain** = *1000000* to do a quick run)
- Click **Generate BEAST file…** and save your file.
  (We saved it as *samlpe1.xml* in *sample* folder)

- Launch ***BEAST v*.exe***
- Enter a **Random number seed** like 12345
- Choose your BEAST XML input file.
  (From our sample files, we would select *sample1.nex* in the Sample folder)
- **Run** it!
- After the program is finished, you will find a *.trees* file in the same folder of your .xml file.
  (In our example, *Sample1 .trees* is our trees data set.)

4.1.2 How to obtain trees data set from PAUP. (Back to Top)

- Use **Lset** or **Pset** command set for ML or MP analysis with option "**Collapse= NO;"**
- Define outgroups and root the trees with **"roottrees OUTROOT=MONOPHYL;"**
- Save all of your trees using "**format=NEXUS**" option.

Important: Trees with polytomies are not accepted by S-DIVA!

4.1.3 How to obtain trees data set from other phylogenetic programs. (Back to Top)

If you are using Mrbayes, it is helpful to define outgroups and specify rooting before you run mcmc. If you did not define outgroups and specify rooting before the MrBayes run, you can root all trees using PAUP or other available software. You can load the MrBayes output file (**.run.t*) as your trees data set once the trees have been rooted.

For other phylogenetic programs, there are two methods for making a trees data set:

Method 1:

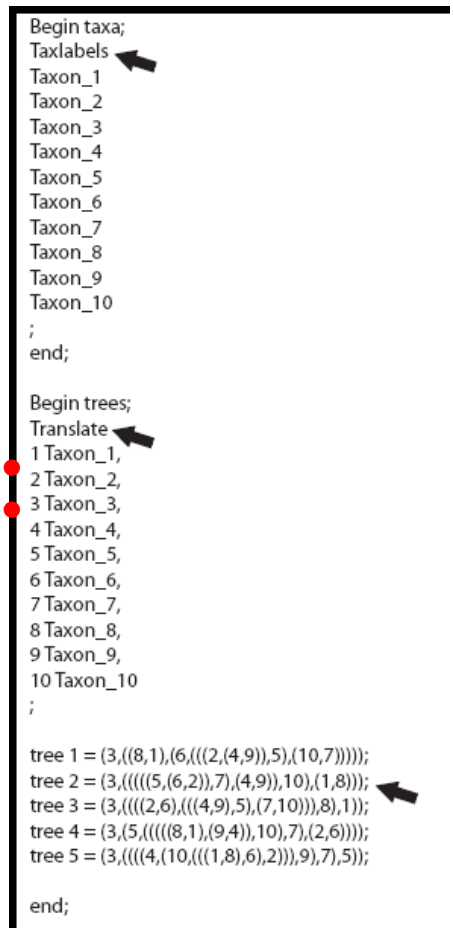
- Save the trees as Nexus format from whatever phylogenetic program you are using.

Method 2:

- Save the trees as PHYLIP format.
- Load the trees into PAUP then export the trees use "**format=NEXUS**" option.

Important: Trees with polytomies are not accepted by S-DIVA!

4.1.4 The bare essentials for the tree file. (Back to Top)



Our sample trees file includes considerable amounts of phylogenetic program output, but here is what a trees file **must** contain. An example, which can be accepted by S-DIVA, is shown to the left.

- Your taxa should have unique labels or names (upper arrow).
- Translate your unique taxon names into integers (1- ~) (middle arrow). These integers are required for the ID field in the S-DIVA program.
- Your file should contain one or more phylogenetic trees in set notation (lower arrow). Many tree generating programs such as PAUP*, MrBAYES, etc. include a lot of extra information within each tree such as branch lengths, likelihood values, and so on. This extra information does not hinder S-DIVA, but it is also not essential.

4.2 How to make a Final tree.

4.2.1 How to make a final tree by BEAST. (Back to Top)

- Launch ***TreeAnnotator*.exe***
- Set **Burnin** and **Posterior Probability**
- Choose your trees data set as Input Tree File

(In our example, we chose *sample\sample1.trees*)

- Choose Output File

(We saved it as *Sample1_Final_Tree.tre* in *sample* folder. *Sample1_Final_Tree.tre* is our final tree.)

4.2.2 How to make a final tree using PAUP. (Back to Top)

- Define outgroups and root the trees with **"roottrees OUTROOT=MONOPHYL;"**
- Export a tree using "**savetrees format=NEXUS**" commands.

4.2.3 How to obtain a final tree from other phylogenetic programs. (Back to Top)

If you are using Mrbayes, you should make the final tree using BEAST or PAUP*.

To make a final tree using other phylogenetic programs, there are three methods for making a final tree:

Method 1:

- Save the tree as Nexus format from whatever phylogenetic program you are using.

Method 2:

- Save the tree as PHYLIP format.
- Load the tree into PAUP then export the tree use "**format=NEXUS**" option.

Method 3:

- Make a text tree file by yourself as the following format:
  tree=(((((6,((2,4),((9,8),3))),1),(7,11)),10),5);

Important: Trees with polytomies are not accepted by S-DIVA!

4.2.4 The bare essentials of the final tree. (Back to Top)

Final tree files may contain various types of commands and information relevant to the program that created them (e.g., #NEXUS, Begin trees;, etc.) An S-DIVA final tree ***must*** contain a line of text that looks like:

tree=(((((6,((2,4),((9,8),3))),1),(7,11)),10),5);

Taxon names must be replaced with integers as shown. The unique integer representing each taxon should be the same as in the trees file. Other types of information in the final tree file may not hinder S-DIVA but are not required.

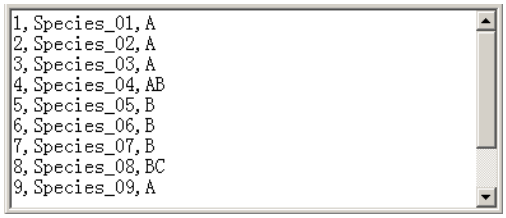
(Tip: What should you do if your consensus tree contains polytomies? **1 –** You can try creating a majority rule consensus tree with compatible groups with less than 50% support allowed. **2 –** Do not use a final tree. Instead you can estimate each node individually using *Estimate a node*.)

4.3 How to make Distributions file

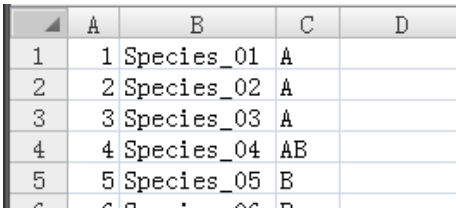
4.3.1 How to make a Distributions file. (Back to Top)

Name the distributions **A, B, C, etc.** and specify multiple-area distributions like **BD or ACE**. Only letters from **A to O** can be used (Ronquist, 1997, 2001).

- Launch ***S-DIVA***
- Select **File > Load Trees** and navigate to your trees data set
- Select **File > Save Distribution** and save it as a .csv file
- Open the your saved .csv file in a text editor or Excel
- Input the distributions after the species name like this:



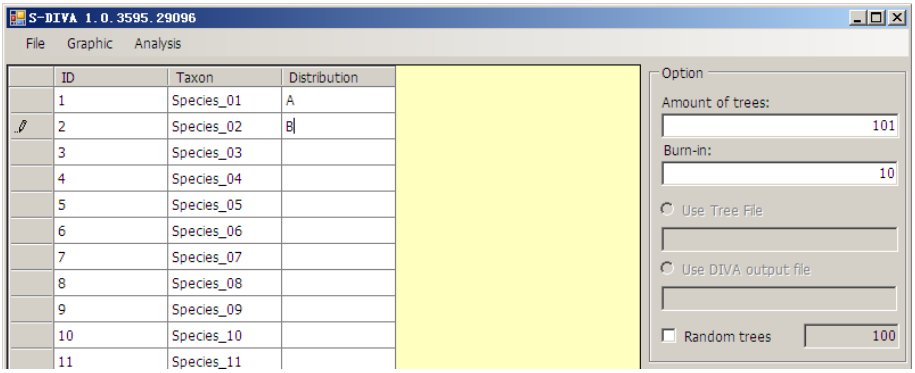
or



4.3.2 How to input distributions in S-DIVA. (Back to Top)

Name the distributions **A, B, C, etc.** and specify multiple-area distributions like **BD or ACE**. Letters from **A to O** must be used (Ronquist, 1997, 2001).

- Launch ***S-DIVA***
- Select **File > Load Trees** and navigate to your trees data set
- Type your distributions directly into the **Distribution** column.
- Remember to save the distributions you have entered to a file for use in future analyses. Save the distributions as a .csv file using **File > Save Distribution**.



4.4 Examples

4.4.1 How to process output file from Mrbayes

- Load your nex file into PAUP
- Import the trees file generated by Mybayes into PAUP:

gettrees file=c:\totaldna.nex.run1.t (replace with your trees file) Unrooted=Yes;

- Define the outgroup:
  outgroup 1 2 3 4(replace with your outgroups) /ONLY;
- Root the trees:
  roottrees **OUTROOT=MONOPHYL**;
- Export the **trees data set**:
  savetrees file='C:\data_set.trees'(replace with your file name) format=NEXUS root=Yes BRLENS=Yes SAVEBOOTP=BOTH REPLACE = YES MAXDECIMALS=0 FROM=1 TO=200001 (replace with the total number of your trees);
- Export the **final tree**:
  CONTREE All /PERCENT=50 STRICT=No SEMISTRICT=No MAJRULE=Yes LE50=Yes GRPFREQ=Yes ADAMS=No INDICES=No SHOWTREE=Yes treefile='c:\final_tree.tre'(replace with your file name) replace=yes;
- 6. Launch S-DIVA. Import your trees data set.
- 7. Rebuild your distributions file. The distributions of outgroups should also be defined.
- 8. Import the final tree into S-DIVA and run analysis.

5. Explanation of messages (back to contents)

1. **Allowing reconstructions will need a larger amount of free space on disk and will take a longer time to run. Are you sure to use it?**
   User will get this message when the number of taxa is more than 64 and reconstructions are allowedsuch that $F(x_n)_t = \frac{i}{D_t}$. Allow reconstruction will need a larg amount of free space on disk and may take a long time to run.
   For example, processing 100 trees with 84 taxon (max areas of each node =4) needs about 5G free space on disk and will take about 2 hours. You may click “yes” to enable “Allow reconstructions” or click “No” to disable it.
2. **Cannot accept polytomies! ...**
   The current version of S-DIVA can handle only fully bifurcating trees. Future releases of S-DIVA may be able to handle soft and hard polytomies. Check your trees for polytomies. You may also see this error message if your trees have not been rooted.
3. **Cannot format the tree!**
   The S-DIVA program accepts phylogenetic trees (a trees dataset and a final tree) generated by the programs BEAST (Drummond and Rambaut, 2006), PAUP* (Swofford, 2003) and MrBayes (Huelsenbeck & Ronquist, 2003). Other tree file formats may cause this error. The error may also occur if you are using an accepted format but have syntax errors in your file (e.g., a misplaced semicolon, a missing "end" command, etc.). Opening the file using PAUP* may help you determine if there is a syntax error, and, if so, what it is.
4. **Cannot process the computed file!**
   There is something wrong with the temp file. Please delete the folder “Temp” and restart the S-DIVA.
5. **Cannot process the trees!**
   There is something wrong with the temp file. Please restart S-DIVA.
6. **Distributions must be labeled using the letters A to O only!**
   Remember that distributions must be labeled using the letters **A** to **O** only and should include no more than 15 unit areas. S-DIVA does not highlight rows containing unrecognized characters.
7. **Distributions of omitted taxon should not be null!**
   When “with omitted taxa distributed in” is checked, you must define the distribution of the omitted taxon.
8. **Distributions should not be null!**
   You have not provided distribution information for one or more taxa. S-DIVA should highlight rows missing distribution information one at a time. If not, check your distribution file or the entry fields in the Distribution column in S-DIVA to see which cells were left blank. You may also prompt this error if your distributions include unrecognized characters.
9. **DIVA12 is found in processes; do you want to end it?**
   DIVA1.2 is running on your computer. You cannot run DIVA and S-DIVA at the same time.
10. **Burn-in error!**
    The value of the Burn-in must greater than or equal to zero but smaller than the amount the trees.
11. **error - no distribution specified**
    Your group of organisms should include no more than 127 taxa. The distributions should include no more than 15 unit areas.
12. **error - no tree specified**
    Your group of organisms should include no more than 127 taxa. The distributions should include no more than 15 unit areas.
13. **error - optimal reconstruction requires too many dispersals**
    The optimal reconstruction requires too many dispersals. You need to decreases the number of taxa or distributions.
14. **Missing DIVA results of tree...! Cannot do the analysis!**
    There is something wrong with the temp file. Please delete the folder of “Temp” and restart the S-DIVA.

15. **Need at least one tree!**
    You need at least one tree in the trees file to run S-DIVA!
16. **Your group of organisms should include no more than 127 taxa!**
    Your group of organisms should include no more than 127 taxa. The distributions should include no more than 15 unit areas.

6. References (back to contents)

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